This support document references the Multiple Zone Concentration Measurement Procedure in appendix C to part 63. We are in the process of amending appendix C to part 63 to add this procedure. A notice proposing to amend appendix C to part 63 will appear in the Federal Register in the near future.

Technical Support Document for the Evaluation of
Aerobic Biological Treatment Units with Multiple Mixing Zones

I. <u>OVERVIEW AND PURPOSE</u>

This document is intended to provide information to assist anyone who needs to evaluate the performance of a biological treatment unit that does not meet the definition of an "enhanced biological treatment system or enhanced biological treatment process" (not considered a "thoroughly mixed treatment unit") because of limitations in overall unit mixing. This document is intended as support for evaluation of biological units with multiple mixing zones. The evaluation of the biological treatment unit can be used for certain compliance demonstration provisions in connection with Appendix C of 40 CFR part 63. Potential users of this document include owners and operators of sources who must demonstrate compliance with the requirements for biological treatment units presented in Appendix C of 40 CFR part 63, as well as enforcement personnel evaluating whether a specific biological treatment process meets the performance criteria required for regulation compliance. It is therefore assumed that readers of this document are familiar with the requirements of Appendix C of 40 CFR part 63, and consequently those requirements are not restated in this document. Users of this information should be familiar with conventional techniques for evaluating the extent of mixing in a biological treatment unit. This information is intended for clarification purposes only, does not constitute final agency action, and cannot be relied upon to create any rights enforceable by any party.

The purpose of this document is to provide technical support and procedures to determine the performance of a biological treatment unit that does not meet the criteria for being considered a "thoroughly mixed treatment unit" within the meaning of the enhanced biological treatment process definition in 40 CFR 63.111. The objectives of these evaluation procedures are to evaluate the performance of a unit that does not quickly disperse the entering wastewater and recycled biomass throughout the unit due to the design and operation of the unit. The evaluation of the effect of mixing limitations would provide an assessment of the volatilization of the compounds of concern as well as the biodegradation rates of those compounds.

Several alternative approaches are presented for evaluating the performance of a biological

treatment process that is not considered to be "thoroughly mixed". All of these procedures are considered to provide equally acceptable assessments and no one procedure is considered to take precedence over another.   In some cases, however, it is not possible to use some of the procedures because of site specific conditions. These evaluation procedures have been designed to allow, to the extent reasonable, the use of existing information and to minimize the amount of new information that is required to evaluate the mixing characteristics of your system. After implementing the procedures of choice, it is necessary for you to have defined zones that have substantially uniform characteristics, especially the concentrations of volatile organic compounds. It is therefore recommended that in those cases where sufficient information is not available to successfully define zones using existing information, you should consider developing additional information to define zones with substantially uniform characteristics.

Some of the guidance provided in this document may not be needed for each procedure described in this document. For example, a laboratory based procedure may require uniform dissolved oxygen concentrations within a zone, whereas the multiple zone concentration measurement procedure may not require uniform dissolved oxygen concentrations. In other methods that require a characterization of the biological process, the concentrations of dissolved oxygen concentrations can be important.

II.   BACKGROUND

Guidance for the evaluation of whether a biological treatment unit is a "thoroughly mixed treatment unit" is provided in the document **Technical Support Document for Evaluation of Thoroughly Mixed Biological Treatment Units** (11/98). This document defines procedures that may be used to divide a biological treatment unit into two or more mixing zones, with each mixing zone potentially considered a "thoroughly mixed treatment unit". The mixing zones approach presented here is different from a tanks in series approach, because there is substantial exchange of material among the different mixing zones. This exchange of material among the different mixing zones is characterized by the concept of a recycle ratio that is applied to each of the interacting zones. The more general computer modeling approach that accounts for exchange of

material among the different mixing zones is described in section H. Tables 5 and 6 address methods of estimating the extent of backmixing.

III.   DESIGN CHARACTERISTICS THAT INFLUENCE THE REQUIREMENTS FOR MULTIPLE MIXING ZONES

This section describes the characteristics of units that are considered to contribute to multiple mixing zones in biological treatment units. The presence of multiple mixing zones is of concern because of the potential of volatilization as opposed to optimum biodegradation in some of the entrance zones.

Biomass separation and agitation are two important characteristics that influence the performance of a biological treatment system. The biomass characteristics can be different in the different zones in a multiple zone system. The uniformity of the biomass characteristics can be improved in a system that is designed or operated so that biomass separation occurs exterior to the aeration system (e.g., secondary clarifier with return of separated biomass to the aeration unit), with return of the separated biomass to the inlet of the system. In the design of the multiple zone system, the unit may have segments that have little or no observable agitation (quiescent zones in the air emission models), or segments with uneven liquid flow patterns, both in direction and velocity. Even with the presence of relatively stagnant zones, there should be enough fluid flow in each mixing zone to support the biomass suspension in the water column. Symptoms of a failure to support biomass in the water column include biomass layers, low dissolved oxygen or anaerobic decay at the base of the floor in these zones, and less overall biomass generation in the system than is theoretically expected. If biomass is not removed from the system with a clarifier, continued accumulation of the biomass in the system will require removal by dredging if the biomass is not removed by degradation in the biomass layers. The presence of biomass settling does not preclude the use of that section of the basin in the calculation of HAP removal, but the presence of biomass settling is an indicator that sections of a basin with substantially different biomass concentrations should be modeled as separate zones.

Baffles reduce mixing in the unit as a whole and the presence of internal baffles suggests deliberate control and restriction of mixing. Baffles can be intentionally included when designing a system with multiple mixing zones. The absence of baffles does not indicate the absence of multiple mixing zones.

One potential indicator of the need for the use of multiple mixing zones is a high length to width ratio in the treatment unit. Mixing in biological treatment units depends on the length to width ratio, the dispersion characteristics, and the retention time in the reactor. Long units are more difficult to mix uniformly. Generally, a length to width ratio of four to one, or greater, is considered a high ratio.   Vivona (1983) states that plug flow sizing would be based on a length to width ratio of 4:1 to 12:1[1] . The requirements for multiple mixing zones can be much less than the requirements for plug flow design. In the technical approach described here, plug flow characterization requires 10 well-mixed zones (10 zones), and the characterization for multiple mixing zones is restricted to 2 to 5 zones. Additional information about well-mixed reactors, multiple reactors in series, and plug flow is described in Levenspiel[2], and Bailey and Ollis[3].

Multiple mixing zones are used to characterize large aeration basins. These large basins may be represented as a group of interacting zones. In a large aeration basin, these multiple zones may be required to account for differences in the component concentrations, in the biomass characteristics, and in the aeration characteristics.

 Aeration that is greater near the inlet of the unit suggests a design for multiple mixing zones that do not have the same conditions in each zone. The greater loading in the initial zone could cause a greater oxygen demand near the inlet. This would imply that the inlet loading is not distributed throughout the unit and significant volatilization may occur prior to efficient biodegradation. The presence of non-uniform agitation and other characteristics such as concentrations of chemicals and concentrations of biomass do not imply that the unit does not meet the requirements for acceptable biodegradation performance, only that special procedures should be followed to evaluate the biodegradation performance.

Quiescent zones separating agitated zones may or may not be well-mixed . For example, surface units may be considered well mixed and uniform within the agitation zone around each surface aerator, but the aeration unit as a whole may not be well mixed throughout the entire unit. In dividing a unit containing multiple aerators into mixing zones, the zone definition should not be smaller than the zone around an aerator in a surface agitated basin that is uniformly agitated with surface aerators. Units designed so that the wastewater flows sequentially from one aeration unit to another may be considered as multiple mixing zones with one mixing zone for each aerator in the path of wastewater flow through the unit. This flow in series may be determined by inspection, or by tracer testing, or by design and operating characteristics.

Examples of design features that may result in poor biodegradation of the compounds in the entering wastewater in the entrance zone of a multiple mixing zone unit include (1) the absence of quick dispersion and thorough mixing and (2) the potential for significant volatilization prior to biodegradation. These two factors are interrelated in that quick dispersion and thorough mixing must occur prior to significant volatilization of the compounds of concern for the system to achieve efficient destruction through biodegradation. Certain design characteristics may lead to problems with respect to these factors. Some of these factors are discussed in the following sections.

IV.   GENERAL PROCEDURES FOR EVALUATING THE MIXING CONDITION OF A BIOLOGICAL TREATMENT UNIT

A.   *Overview of procedure*

This section presents a list of procedures that can be used to evaluate the mixing conditions of a biological treatment unit that has multiple mixing zones. The overall performance of the biological treatment unit is characterized by three factors: the fraction of the compounds entering the unit that is biologically degraded, the fraction of the compounds that is emitted from the unit as air emissions, and the fraction of the compounds that remains in the wastewater after treatment in the unit. If the total removal by biodegradation is acceptably great for the entire

treatment unit, the unit may be considered as an acceptable biological treatment process for the purpose of regulatory compliance. In some cases, there may be very aggressive biodegradation and low stripping in the first part of the biological treatment unit. If the required destruction of compounds is achieved in that first part of the biological treatment unit, the characterization of the other parts of the biological treatment unit would not be required. If you choose to only characterize a section containing multiple zones of a large aeration basin, you should account for the internal recycle effects at the end of the section, because there will be backflow from outside the section back into the zone at the end of the section.

The first step is to subdivide the unit into a series of zones that have substantially uniform characteristics within each zone, such as organic compound concentrations, dissolved oxygen concentrations, and biomass concentrations. Then, the zone that can be considered as a well-mixed flow entrance zone is identified. If multiple inlets of wastewater are present, two or more entrance zones may be present. Depending on the unit, an entrance zone could extend for as much as one half the volume of the system. The procedures for evaluating the number of mixing zones are described in Section C and these procedures can be summarized as identifying zones that have uniform conditions and concentrations of components. The division of the system into zones depends on the complexity of the system and the technical approach. If laboratory based measurements of the biorate constant are used, it is important to match the dissolved oxygen and other important variables in the laboratory with those same important variables in the full scale system. With other procedures, the dissolved oxygen concentrations are less important. One of the procedures relies primarily on evaluations of the concentrations of the compounds of interest and the aeration characteristics: with that procedure, it is important to select zones with substantially uniform compound concentration and agitation characteristics.

The second step is optional and can be used to reduce the resources required for regulatory compliance if biological rate measurements are used to characterize the performance of the system. If the emission potential for the well-mixed flow entrance zone is greater than or equal to the other mixing zones, then only the first zone is evaluated and the performance of the

other zones are assumed to be equal to the first zone. When the performance of the overall unit is evaluated by this approach and determined to be acceptable by this method, then an evaluation of the remaining zones are not required. The key to the confidence in this approach is the assurance that the first zone does not have superior performance to the remaining zones (the ratio of biological removal to air stripping is not greater in the first zone). Design factors that could prevent the use of this optional procedure include more aggressive biodegradation in the initial zone due to special biological activity from the recycled biomass, less aggressive aeration in the initial zone, and deeper unit depth in the initial zone.

The third step in the evaluation process is to identify the number of mixing zones that are needed to evaluate the system (2,3,4,5, or a maximum of 10) and proceed with the appropriate form for the number of mixing zones. The number of mixing zones that are needed to evaluate the system can be less than the total number of zones that are identified in step 1. Large aeration basins can have more than two dozen surface aerators that could theoretically be considered as a separate zone for each aerator, but due to the mixing characteristics, four or fewer zones could be selected for evaluation purposes. In this case several aerators would be included in a single zone. Procedures to identify the characteristics of mixing zones are described in Section C of this document, and forms are provided to complete the appropriate calculations for this identification of the number of mixing zones.   The three procedures are design evaluation, tracer studies, and in−basin measurements. All of these procedures are considered to provide equally acceptable assessments of the number of mixing zones and no one procedure is considered to take precedence over another. Selection of the procedure will depend on the availability of information, the relative ease of obtaining the necessary information, and/or personal preferences.

If there is a question about how many mixing zones that should be used for describing the unit, use more zones rather than less. If additional zones are used to characterize a basin, the recycle ratios should be appropriately adjusted. Some of the technical approaches do not require the evaluation of recycle ratios.

Under some special design conditions, the overall unit cannot be considered to be either well-mixed, multiple-zones, or plug-flow. There are several different procedures for evaluating units in this document and all of the procedures in this document may not apply to those systems with specials designs, due to abnormal flow conditions, poor suspension of biomass, uncharacterized dissolved oxygen gradients, or other special site-specific factors. For those systems with special design conditions, the use of some of the procedures in this document may require detailed modeling of the actual site based upon appropriate modeling techniques, using the methods provided in this document as general guidance.

B.   *Determination that the Unit is Not Well-Mixed*

The first step in the general determination of the biological performance of a wastewater treatment unit is to determine that the unit can not be considered as well-mixed. If an initial evaluation of the procedures in the document **Technical Support Document for Evaluation of Thoroughly Mixed Biological Treatment Units** indicates that the there is a likely probability that the unit would not be considered well-mixed, then proceed to the evaluation of the multiple mixing zones.

C.   *Determination of the Number of Mixing Units*

1. Initial mixing zone

When you break an unit into zones, one or more of the zones is an initial mixing zone. You may determine that the unit has an initial mixing zone that can be considered as well-mixed by design evaluations, by tracer testing, by concentration testing, or by initial inspection. If the definition of the initial mixing zone cannot be considered as uniform or well-mixed, you should redefine the initial mixing zone so that it can be considered to be substantially uniform in conditions. Also, sampling of the initial mixing zones should be carried out in a central position in these zones so that the measured concentrations are representative of the conditions throughout the mixing zone.

2. Number of mixing units from dispersion analysis

In the case of submerged aeration, if you have a spiral flow aeration systems you may use Form 15 to estimate the dispersion coefficient by the method of Fugii or you should use the default value of 0.068 m^2/s (Chambers) for the other types of submerged aeration systems. Next, use Form 16 to Calculate the value of u and L from the mean velocity and length of the aeration unit; then, use those values to calculate the dispersion number (D/uL). Use Table 1 and Table 2 to select the number of mixing zones from the value of the dispersion number. The number of units by this method is the equivalent number of tanks in series that will represent the characteristics of the dispersion and may be somewhat conservative when compared to other methods. The following equation describes dispersion in a closed system.[4]

$$\sigma^2 = 2 \, (D/uL) - 2 \, (D/uL)^2 \, (1 - e^{-uL/D})$$

Table 1 presents some of the calculated values of the dimensionless variance using the above relationship.

Table 1. Relationship between the dispersion number and the dimensionless variance.

σ^2	D/uL
0.9674836	10
0.9216251	4
0.8975636	3
0.8522453	2
0.8155969	1.55
0.7990033	1.4
0.7652601	1.16
0.6867261	0.8
0.654858	0.7
0.6159904	0.6
0.6026241	0.57
0.5676676	0.5
0.5198208	0.42
0.4992198	0.39
0.4769162	0.36
0.4264213	0.3
0.3772895	0.25
0.332554	0.21
0.240831	0.14
0.1958027	0.11
0.1638002	0.09

The number of tanks in series model may be used for systems with either subsurface aeration or surface aeration basins.

The dimensionless variance σ^2 is then related to the number of tanks in series (no back mixing) with an equivalent variance, where the number of mixing units[5] equals the reciprocal of the dimensionless variance, $1/\sigma^2$. The number of tanks in series that corresponds to the dimensionless variance depends on the extent of back mixing. The amount of back mixing in the tanks in series model is defined by the recycle ratio. The internal recycle ratio is the ratio of the flow due to mixing in the unit toward the inlet to the flow in the wastewater plus any external recycle. The recycle ratio in basins with surface aerators are estimated to be in the range of 2 to 4. The recycle ratio may be estimated from the local basin flow rates if they are available. If the

backflow from zone N+1 to zone N is measured as 3 m^3/s and the flow rate of the wastewater to be treated plus the recycle flow is 1 m^3/s, the recycle ratio is estimated as 3/1 or 3.   The recycle ratio is used with a number of tanks in series to model the mixing characteristics of the actual unit. Since the mixing characteristics of the actual unit are generally not identical to the theoretical mixing characteristics of the tanks in series with recycle model, the success of the model in describing the actual unit may depend on the selection of the number of zones and appropriate values for the recycle ratio. The appropriate value of the recycle ratio may depend on the selection of the zones. Information on the dispersion and flow in the system should be used to estimate the value of the recycle ratio.

The internal recycle in the tanks in series model is the flow rate of tank N+1 back to tank N. Figure 1 illustrates the model that was used to calculate the dimensionless variance for use in Table 2. The values in Table 2 were calculated and rounded to two significant figures.
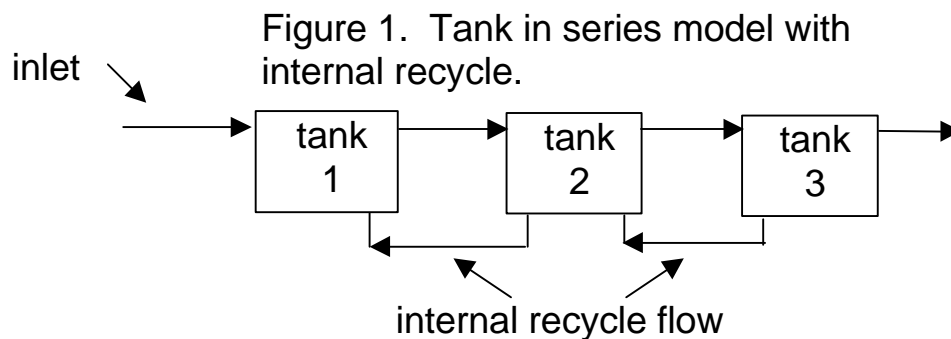

Figure 1. Tank in series model with internal recycle.

tanks	Internal recycle ratio				
	0	1	2	3	4
1	0.99	0.99	0.99	0.99	0.99
2	0.467	0.78	0.86	0.85	0.88
3	0.33	0.62	0.76	0.81	0.86
4	0.244	0.52	0.66	0.75	0.81
5	0.193	0.44	0.59	0.68	0.74
6	0.16	0.38	0.53	0.63	0.66

Table 2. Calculated dimensionless variance numbers σ^2 for various number of tanks in series at specified internal recycle values

To use Table 2, identify the applicable column that corresponds to the recycle ratio identified for the unit of interest. Look down the applicable column to locate the row containing the dimensionless variance that was estimated. The corresponding number of tanks in series is listed in the left column of that row. Linear interpolation may be used for Table 2.

Table 3 may be used for an assumed default recycle ratio of 3 for the biological treatment system with estimated dispersion numbers.

Table 3. Default values for the number of mixing units based on estimated dispersion numbers.

Dispersion number, D/uL	Number of mixing units
D/uL > 10	2
10 > D/uL > 1.4	3
1.4 > D/uL > 0.7	4
0.7 > D/uL > 0.5	5
0.5 > D/uL > 0.42	6

3. Number of mixing units from tracer analysis

You should only interpret the mixing characteristics of your unit by tracer analysis if you

are experienced in tracer testing and understand the complexities of tracer interpretation. The following discussion presents an overview of the use of tracer testing to characterize the mixing characteristics of a unit.

When a sample of tracer is instantaneously added to the inlet pipe of a biological treatment unit, the amount of tracer leaving the unit in the exit pipe is measured as a function of time. The tracer measurements may be analyzed to determine the mean residence time and the standard deviation of the distribution. The exit tracer concentration will increase with time, reach a peak or maximum concentration, and then decay with time. Other observations of interest include the time for the maximum in the peak and the absence of multiple peaks. You must correct the results of the tracer analysis for recycle flow systems, because some of the exiting tracer will be returned to the unit with the recycled sludge. If the hydraulic residence time (volume divided by inlet flow) is approximately equal to the tracer residence time, this is an indication that the selection of tracer was good and that the unit does not have significant bypassing and abnormal flow patterns[6]. The ratio of the standard deviation to the mean residence time is the dimensionless variance. Look up the equivalent number of mixing units from the measured dimensionless variance in Table 4 if your unit has a recycle ratio of 2-4. The equations relating the dimensionless standard deviation to the number of units are discussed in the previous section.

Table 4. Default values for the number of mixing units based on measured dimensionless variances obtained from tracer testing. (Based upon a recycle ratio of 2-4)

dimensionless variance, σ^2	Number of units
$\sigma^2 > 1$	2
$1 > \sigma^2 > 0.8$	3
$0.8 > \sigma^2 > 0.66$	4
$0.66 > \sigma^2 > 0.57$	5
$0.57 > \sigma^2 > 0.53$	6
$0.53 > \sigma^2 > 0.47$	7
$0.47 > \sigma^2 > 0.41$	8
$0.35 > \sigma^2$	10

Tracer testing can provide information that may be helpful in evaluating the number of tanks in series and the internal recycle ratios needed to use the tanks in series model with backmixing. The peak or maximum tracer concentration discussed in the previous paragraph may be used to characterize the unit. The dimensionless peak time is the time of maximum tracer concentration at the exit of the unit divided by the hydraulic residence time of the unit. The dimensionless peak concentration is the maximum tracer concentration at the exit of the unit divided by the ratio of the amount of tracer to the volume of the unit. Tables 2, 5, and 6 may be used to select the number of tanks and internal recycle ratios for unit evaluation.

Table 5. Dimensionless peak times at various internal recycle values					
tanks	Internal recycle ratio				
	0	1	2	3	4
1	0.005	0.005	0.05	0.05	0.05
2	0.5	0.43	0.37	0.33	0.3
3	0.66	0.53	0.47	0.434	0.4
4	0.74	0.6	0.54	0.5	0.46
5	0.8	0.65	0.58	0.54	0.51
6	0.83	0.68	0.61	0.57	0.54

Table 6. Dimensionless peak concentrations at various internal recycle values					
tanks	Internal recycle ratio				
	0	1	2	3	4
1	0.997	0.997	0.997	0.997	0.997
2	0.740	0.550	0.440	0.363	0.309
3	0.819	0.627	0.517	0.442	0.386
4	0.907	0.68	0.573	0.498	0.443
5	0.991	0.730	0.618	0.543	0.487
6	1.07	0.777	0.656	0.580	0.524

4. Number of mixing units from design factors

Some units can have screens, baffles, and flow pattern designed to promote a controlled path of wastewater through the unit, rather than general mixing. For those cases, it may be

appropriate to separate the unit into zones based upon the physical construction of the unit. If the unit contains nonuniform agitation or nonuniform aeration, zones should be selected that have relatively uniform surface characteristics. The primary guidance for the selection of the number of mixing zones in this case is that too many units will not adversely affect the results, but too few can adversely affect the accuracy of the unit evaluation.

5. Number of mixing units from measurements of concentrations
The number of mixing units may be obtained from measurements of concentrations of volatile compound concentrations at multiple locations in the unit. Zones are selected based upon these concentrations and a zone does not need to have the same concentration throughout the zone. Emissions from an area within the zone that are higher than the average for that zone can be offset by lower emissions from other areas in that same zone that are lower than the average if the concentrations in the zone are substantially uniform. In general, the division of the unit into more zones will increase the accuracy of the estimated air emission rate from the unit, but this increase may be very little for some systems. For systems with a continuous change in concentration across the surface of the system, a 15% difference in the volatile compound concentrations from the average value in a zone could be considered substantially uniform for the purpose of these calculations (range of approximately 30% of the mean). A consideration of the impact of the number of zones on the estimated fraction biodegraded and the estimated air emission rates can help resolve issues in the determination of the number of zones. A larger difference from the mean can be used if it can be shown that the zone size is sufficiently small such that numerical errors introduced by the larger grid size are an acceptably low value.[7] In some cases, errors in the grid size are not important in the evaluation of a biotreatment unit:

- the biodegradation removal effectiveness (f_{bio}) is substantially greater than required for regulatory compliance,

- a decrease in the grid size has no significant impact on the biodegradation removal effectiveness, and

- the biodegradation removal effectiveness is not sufficient for regulatory compliance and additional accuracy would not change the evaluation.

In other cases, errors due to a larger grid size can be important in the evaluation of a biotreatment unit: the biodegradation removal effectiveness (f_{bio})is neither significantly greater than or significantly lower than the value required for regulatory compliance. In this case, improved accuracy that is thought to be associated with a smaller grid size may be more effective in resolving uncertainty in regulatory compliance issues.

For example, consider a biotreatment unit with a requirement that 90 percent of the inlet HAPs be biodegraded (f_{bio}). If data collected during an initial performance test show that the unit typically achieves an f_{bio} of 91 percent, the zone size should be selected such that the numerical error introduced by using fewer zones is no more than 1 percentage point. However, if the initial performance test data show that the unit typically achieves an f_{bio} of, say, 97 percent, an acceptable numerical error may be 2 to 3 percentage points.

D.   *Determination of the Relative Performance of the Initial Mixing Unit*

The second step is a determination of whether the initial mixing zone has an equal or greater emission potential than the other zones. This step is optional and can be used to reduce the resources required for regulatory compliance. If the emission potential for the well-mixed flow entrance zone is greater than or equal to the other mixing zones, then only the first zone is evaluated and the performance of the other zones are assumed to be equal to the first zone. When the performance of the overall unit is evaluated by this approach and determined to be acceptable by this method, then an evaluation of the remaining zones are not required. If the required biodegradation is achieved in the initial mixing zone, an evaluation of the remaining zones is not required.

You must provide an assurance that the first zone does not have superior performance to the remaining zones if you use this option. The first zone is generally the zone of the most environmental concern since the concentrations are greater, the relative biorates are potentially less, and mixing at the entrance is a potential problem. You should consider any design factors that could prevent the use of this optional procedure. Other design factors could include a deeper unit depth in the initial zone, the location of the inlet wastewater, and special mixing characteristics near the wastewater conduit.

Another factor that could conceivably prevent the use of this option include more aggressive biodegradation in the initial zone due to special biological activity from the recycled biomass. It has been suggested that the biomass has a greater potential for active enzyme formation and lower concentrations in the biomass, resulting in strong initial uptake of concentrations by the biomass.

If there is less aggressive aeration in the initial zone than in other zones, the initial zone could conceivably have a lower rate of stripping than in other zones. This could be especially important for surface aerator units and for submerged aeration units with uneven aeration.

E.   *Determination of the Overall Unit Performance from the Performance of the Initial Mixing Unit*

The fraction of the concentration loading that is removed as air emissions and as biological products is estimated from the use of Form 3 using the measured biorate with the concentrations of compound and biomass in zone 1 and the characteristics of zone 1. These same fractions are then applied to each of the other zones in the biological treatment unit in sequence from zone 2 to the last zone of the unit. The fractions of removal by biodegradation and air emissions are estimated as follows:

$$f_{e,i} = f_{e,1} f_{r,i-1}$$

$$f_{b,i} = f_{b,1} f_{r,i-1}$$

$$f_{r,i} = f_{r,i-1}\left(1 - f_{e,1} - f_{b,1}\right)$$

The subscript i refers to the fraction in zone i, were i varies from 1 for the first zone to n for the nth zone. If the biological treatment does not have an acceptable biological removal effectiveness by this method, this does not mean that the unit is unacceptable. It does mean that the unit is required to be evaluated by step 3 before it may be accepted as being biologically effective.
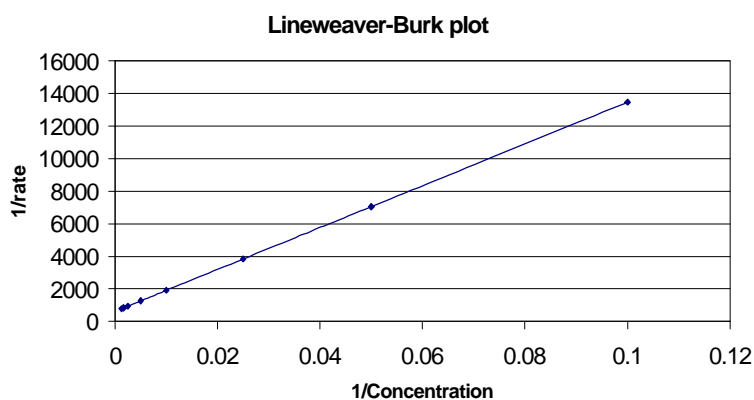
F.   *Determination of the Characteristics of the Mixing Units*
   1. Biological rates

The biological rates are measured in each mixing zone, where required for the procedures in this document. In step 2 only the biological rate (as determined by the appropriate methods in Appendix C of 40 CFR part 63) in the first mixing zone is required. If three or more mixing zones are evaluated, the biological rates for three zones can be measured, and the results plotted by the method of Lineweaver-Burk[8], yielding a straight line with a slope that is related to the first order rate constant and the intercept that is related to the zero order rate constant. The Monod equation then may be used to estimate biorates in zones other than the three that were used to measure biorates. For the purpose of the evaluation the actual biorate is used (gm/L-hr) for each zone.

The following illustration indicates the Lineweaver-Burk method of plotting the Monod rate data to obtain a straight line. Either the data from the correlating straight line may be used for the estimate of the biorate, or the Monod parameters may be determined from the slope and intercept. Formal statistics may be used to estimate the uncertainties in the values of the slope and intercept obtained from this approach.

In some units, you will not be able to use the Monod equation to correlate your data because your system may be more complex. For those systems a different correlation may be used to estimate the biorates for some of the zones. The general guidance for this case is that enough measurements must be carried out to establish an unambiguous correlation. Measurement in the initial mixing zone is always required, and measurement of the last mixing zone is desirable, unless the concentrations are too low for measurement. The user shall find a kinetic model to extrapolate measured biorates to zones that have concentrations that are too low for biorate measurement. The kinetic model that is used should describe the kinetics for the compound of interest that was measured for those mixing zones with higher concentrations.

Lineweaver-Burk plot

2. Submerged aeration rates

Determine the submerged aeration rates for each mixing zone. This procedure is especially critical for systems that may have uneven aeration, either by design or by mechanical malfunction (broken headers, clogged exits). This procedure is less critical if the submerged aeration rates are generally uniform across the entire unit. The following two examples illustrate how the

submerged aeration characteristics can be used to define the mixing zones in the unit.

Example 1.   The treatment unit is a long rectangular channel with two different aeration zones, a higher initial aeration zone, and a lower secondary aeration zone. The unit was divided into four mixing zones on the basis of concentration measurements along the length of the unit. The first two mixing zones are characterized as higher aeration, and the last two mixing zones are characterized with the lower aeration rates.

Example 2.   The treatment unit is a circular tank with flow inlets and exits at opposite sides of the tank. There is a broken header in the center of the tank with heavy aeration.   The unit was divided into three mixing zones on the basis of the observed surface disruption due the broken header. The first and the last mixing zones are characterized with the design aeration rates as confirmed by flow measurement, and the center mixing zones was characterized with the higher aeration rates due to the broken header.

3. Mass transfer coefficients

    You should select the number of mixing units to match the surface aeration pattern, for the presence of non-uniform surface agitation. If there is a grouping of two or more aerators such that their areas of agitation are sufficiently close together that the zone can be considered thoroughly mixed, this grouping can be considered as a set. If an impoundment has 4 of these sets of aerators between the entrance and the exit, four aeration zones could be an initial choice for zones. For more complex situations, mixing zones with different mass transfer coefficients may be required.

4. Biomass concentration

    You should measure the biomass concentrations at several different places in each zone to establish that the biomass can be considered to be uniformly distributed within each mixing zone. If the system is operated with non-uniform biomass concentrations, non-uniform oxygen

concentrations, or non-uniform compound concentrations in a zone, all of the evaluation procedures presented in this document should not be used , and appropriate site-specific methods may be required for some of the evaluation procedures. For abnormal operation, the worst-case measured conditions may be used with the guidance in this document to provide additional assurance that the performance of the unit is acceptable. If this worst case option is used, enough measurements should be taken to reasonably establish the worst case condition. Procedures that rely only on measured concentrations and estimated mass transfer coefficients do not require detailed measurements and characterizations of biomass concentrations and dissolved oxygen concentrations.

5. Dissolved Oxygen Concentration

The same general considerations apply to measurements of dissolved oxygen as with biomass, except that it is much easier to measure dissolved oxygen than biomass, since a dissolved oxygen probe can provide instantaneous measurements. It is possible therefore to make many dissolved oxygen measurements in a mixing zone demonstrating uniform conditions, and therefore potentially reducing the number of biomass concentration measurements that may be required. For effective aerobic biodegradation, the dissolved oxygen concentration will be significantly less than equilibrium (generally less than 7 ppm) and greater than 1.5 ppm (very low dissolved oxygen is an indication of less effective aerobic biodegradation. If the concentration of dissolved oxygen in a zone is less than 1.5 ppmw, the kinetic characterization of the biodegradation in that zone may indicate differences from the kinetic characterization in other zones that have concentrations of dissolved oxygen that are greater than 1.5 ppmw. A minimum dissolved oxygen concentration for aerated stabilization basins should be 0.5 ppmw. The actual dissolved oxygen concentrations that are representative of each zone are used in any laboratory measurements of biodegradation rates.

G.   *Sampling Methods and Locations*

In the initial characterization of the mixing characteristics of a unit, sampling of the water

in the unit is important for an accurate characterization. Some of the methods to characterize the performance of the unit require the measurement of one or more component concentrations at several sampling locations within the unit. The minimum number of sampling locations required to characterize component concentrations within the unit includes (1) the inlet, (2) within unit near the inlet, (3) the exit of the unit, and (4) within center of each mixing zone. Additional sampling locations are initially required to establish the number of mixing zones and to establish that the sampling location is characteristic of the zone. The inlet is sampled directly before entering the particular unit, and the exit is sampled directly at the outfall of the particular unit. The inlet sample may be collected upstream provided conveyance is by closed pipe and no additional streams are added to the conveyance system**.**

The sample within the first mixing zone will be taken as described in the following: first determine the physical dimensions of the first mixing zone.   Sample within the center part of the first mixing zone. Additional samples of the aeration unit contents nearer the inlet should also be taken near the reactor inlet to confirm that the first mixing zone was appropriately chosen. The success of sampling the unit with this method depends on an accurate sampling of the inlet stream after it mixes with the aeration unit contents. Sampling in the unit should be conducted in the flow path of the inlet stream after the inlet flow has an opportunity to mix with the unit contents. The lesser value of either ½ the distance to the closest aerator, a distance of 10 times the diameter of the wastewater inlet pipe , or 10 meters may be used as the maximum sampling distance from the wastewater inlet.

Additional samples will be collected within each additional mixing zone as required by the procedures, and as required for biological rate testing.

1. Collection and handling of samples.

Sufficient grab samples to characterize the concentrations of target compounds should be collected from each of the following locations: (1) the influent to the biological treatment unit; (2) the effluent from the biological treatment unit; (3) the inlet to the aeration unit within the

maximum sampling distance; and (4) near the center of each other relevant mixing zone. The number of samples required to characterize the unit depends on the complexity of the unit and the variability of the inlet waste concentrations and inlet flow concentrations. The relevant mixing zone samples shall be taken anywhere practical within the center of the mixing zone avoiding edge, bottom, or surface effects. When you sample to determine the number and location of the zones, samples are taken in different regions of each zone to evaluate the variability. Note: these samples may be collected by personnel from the sides of the aeration unit, with the assistance of flotation devices, pumps, conduits, and other devices on the sampling equipment to obtain samples that are representative of the center of the mixing zones. Measure the concentrations of the compounds of interest, the biomass, and each characterization parameter (dissolved oxygen, pH, COD) at each of these relevant locations. The aeration unit samples should be collected in the upper part of the basin at a depth of at least 1.0 foot below the surface of the water. When a set of samples is used to characterize the unit, all samples in the unit shall be collected during the same 24 hour period. Each of the sets of samples[1] should be collected to characterize the sampling and unit variability. If more than 3 samples are to be collected for the purpose of zone characterization, then the sample collecting should be carried out at approximately the same time. If time delays are required because of the sampling methods, the sampling locations and times should be scheduled to avoid a bias in the results due to systematic changes in concentrations. The aeration unit samples should be collected during the same time periods that the influent and effluent samples are collected. Under potentially changing conditions of treatment unit operation, samples should be collected for enough days to establish that the operating conditions are stable and that the measured samples are characteristic of those operating conditions.

One method for obtaining representative samples from the zones is to obtain grab samples of the reactor contents removed by a recirculating conduit. Those grab samples should be removed with a zero headspace device, especially if time composite samples are obtained. Samples should be poured from the grab sampling device into sample bottles in a manner that will

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[1]More than 3 samples may be collected from any of the locations, if necessary.

minimize volatilization of organic compounds.   Sufficient hydrochloric acid (HCl) shall be added to each sample to reduce the pH to less than 2 to stop the biodegradation in the sample bottles, unless it is demonstrated that a different pH range is effective for stopping biodegradation and does not cause degradation products present at the lower pH level. The samples shall then be refrigerated at 4° C until analysis.

2. Number of Samples.

When the coefficient of variance[2] for sampling is large, it may be difficult to accurately estimate the mean of the distribution. One method for improving the accuracy of the determination of the mean is to increase the number of data points that is used in estimating the mean of the distribution. The following list presents a recommended minimum number of sequential data points that should be collected from the unit, based upon the measured coefficient of variance.

Coefficient of variance	Minimum number of data points
10	3
15	4
20+	5

3. Measurement of concentrations of relevant compounds.

All sample preservation, storage, and analyses shall be performed in accordance with the NPDES analytical procedures at 40 CFR part 136. You should only use methods that are suitable for measuring the relevant compounds.   All quality assurance/quality control requirements of the applicable method shall be followed.

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[2]The coefficient of variance is the ratio of the standard deviation of the sample mean to the sample mean, multiplied by 100.

4.   Estimation of zone concentrations with limited sampling

In the initial evaluation of the biological treatment unit, detailed sampling may be needed to characterize the performance of the unit. In later evaluations it may not be necessary to collect HAP samples from each zone in a multiple zone biological treatment unit in order to evaluate the overall performance of unit. For example, under the Multiple Zone Concentration Measurement Procedure (appendix C, part 63), the HAP concentration may be estimated for zones located between zones with measured HAP concentration data. The initial unit investigation should provide a sufficient database of measured concentrations in all zones of the treatment unit to allow for interpolation for those zones that are between zones with measured HAP concentrations. The database of HAP concentrations in each zone is developed during the initial biological treatment unit characterization.

The HAP sample collected for the zone should be representative of the average concentration of the zone. However, it is not necessary that the sample be collected at the center of the zone if it has been demonstrated during the initial biological treatment unit characterization that the sample location provides data that are representative of the zone.   Any procedures used to correct the data from the sample location to the average expected concentration of the zone should be developed during the initial biological treatment unit characterization.

H.   *Computer Models*

Computer models may be used to perform the calculations required for step 3. As a requirement for the use of the computer models for the site specific calculations, the following information must be available:

•   an applicable set of site-specific rate data for each relevant compound correlated as Monod constants;

•   a computer program that accounts for concentration variability of the biorate constant with the Monod constants;

• characterization of each mixing zone as a separate unit for modeling purposes, surface agitation effects, submerged aeration, and other factors; and

• evaluation of internal recirculation factors between each mixing zone for use in modeling the recycle streams between each adjacent mixing zone.

The concentrations of the compounds in the mixing zones are available from measurements in the mixing zones of the unit. Concentrations are estimated from the computer model accounting for internal mixing and concentration effects on the biorate. The internal recycle rate and the Monod constants are treated as adjustable parameters, and adjusted until the measured concentrations match the estimated concentrations from the computer model. The computer estimation of the biorates and the air stripping rates are then documented in Form 4, and the overall biological removal effectiveness is evaluated.

I.   *Applicable Multiple Mixing Zone Forms*

Several forms are provided to assist in the organization of information that was used to estimate the biodegradation rates within a multiple mixing zone unit.

1. Form 1. Data Form For The Estimation Of Multiple Zone Compound Fraction Biodegraded And Air Emissions

Form 1 provides a summary of the unit performance (f_{bio} and f_e) based upon measured biorates, measured concentrations, and estimated mass transfer coefficients.

2. Form 2. Data Form For The Estimation Of The Biorate For Each Zone In The Biological Treatment System

This form is used to list the measured biorates from multiple zones and the measured concentrations in each zone. The bioremoval rate constant (sec^{-1}) is calculated from the concentrations and the measured biodegradation rates.

3. Form 3. Data Form For The Estimation Of The Biodegradation Rate For Each Zone

This form is used to estimate the fraction biodegraded and the fraction air stripped in a specific mixing zone of an unit. Form 3 compares the rate of biodegradation for a specific concentration to the rate of stripping for that same concentration. The concentration that is in the zone will depend on the recycle ratios, the number of zones, and other factors.

Form 4. Data Form For The Estimation Of Multiple Zone Compound Concentrations (3 Zones)

Form 4 provides estimated concentrations from measured biorates and estimated internal recycle rates. This form is used to estimate compound concentrations in the zones of units that are characterized by three mixing zones with internal recycle between the units. This kinetic model is different from reactors in series because of the internal recycle causing mixing of zone contents among the three units. This form could be used to confirm the modeling approach or to determine the internal recycling rates for modeling purposes.

5. Form 5. Data Form For The Estimation Of Multiple Zone Biodegradation From Unit Concentrations

Form 5 provides a method to estimate the biodegradation rates of a unit based upon the measured compound concentrations in each unit zone. This method can be useful for the situation in which the compound concentrations are below the detection limits of the measurement method at the exit and near the exit of the unit. The biodegradation rate is estimated as the difference in the inlet loading rate and the sum of the exit removal rate and the air stripping rate. Either forms may be completed or computer models (see Appendix C of 40 CFR part 63) may be used to estimate the mass transfer coefficient in each zone and the actual concentrations in each zone are sampled and measured. Because of uncertainties in the estimation of mass transfer coefficients, this method should not be used when the air stripping rate can potentially account for more than 25 percent of the removal. In the case of steady operation with accurate inlet and outlet concentrations and flows and estimated air stripping rates of a few percent, this method is thought

to provide an accurate estimate of the overall unit biodegradation rate.

V. EXAMPLE FOR THE USE OF MEASURED BIORATES

Example 1 using Form 1 is presented with the forms. This example illustrates how the concentrations and measured biorates in the zones are used to estimate the fraction biodegraded and the fraction of air emissions in the full unit.

Step 1. Identify the number of zones.

Information required by Form 1 is collected from the full-scale unit. Based on tracer testing, three zones are identified for simulating the performance of the full-scale unit.

Step 2. Measure the concentrations in the zones.

Several concentrations are measured for various locations in the three zones that were identified in Step 1. An average concentration for each zone was obtained for use in Form 1. Use the actual measured concentrations, because the concentration in the recycle streams may not necessarily exactly equal the exit concentration from the unit.

Step 3. Measure the biorate for each zones.

The average concentration in each zone was used for measuring the biorates in a bench scale reactor. The biomass from the zone was used in the bench scale reactor. The reactor conditions were adjusted to duplicate the actual zone conditions, including dissolved oxygen concentration, waste concentrations, pH, and temperature.

Step 4. Complete Form 1.

Form 1 is completed and the following are calculated: the fraction biodegraded, the fraction of air emissions, and the fraction remaining in the full unit.

Step 5. Review the results of Form 1.

The fraction predicted that is remaining in the full-scale unit (Form 1, line 23) is compared to the calculated fraction remaining (Form 1, line 13). The concentration in the effluent is compared to the concentration in the last zone. Based upon the data analysis of Form 1 it is concluded that three zones are sufficient to model the full-scale system. If additional zones are needed, the concentrations obtained in Step 2 are used to define additional zones.

VI.   REFERENCES

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